

#### **Cover Page**

BA	RD	<b>Project</b>	<b>Number:</b>	IS-3555-04
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Date of Submission of the report: 18 March 2008

**Project Title: Goats for Maximal Efficacy of Brush Control** 

<u>Investigators</u> <u>Institutions</u>

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**Keywords** *not* appearing in the title and in order of importance. Avoid abbreviations. **Targeted grazing; tannin; terpene; chemical defense; Near infra-red spectrometry.** 

**Abbreviations commonly** used in the report, in alphabetical order:

DM, Dry Matter; NIRS, Near Infrared Reflectance Spectrometry; PEG, Polyethylene glycol;

**Budget:** IS: \$184,600 US: \$155,400 Total: \$340,000

Signature Signature

Principal Investigator Authorizing Official, Principal Institution



# **Publication Summary (numbers)**

	Joint IS/US authorship	US Authors only	Israeli Authors only	Total
Refereed (published, in press, accepted) BARD support acknowledged	3			3
Submitted, in review, in preparation	7			7
Invited review papers				
Book chapters	2			2
Books				
Master theses		1	2	3
Ph.D. theses			1	1
Abstracts	3	1		3
Not refereed (proceedings, reports, etc.)				

**Postdoctoral Training:** List the names and social security/identity numbers of all postdocs who received more than 50% of their funding by the grant.

**E.S. Campbell, Ph. D**., ID 701002075

**Cooperation Summary (numbers)** 

	From US to Israel	From Israel to US	Together, elsewhere	Total	
Short Visits & Meetings	1	2		3	
Longer Visits (Sabbaticals)					

#### **Description of Cooperation:**

Only four teams in the world currently have fecal NIRS methodologies to elucidate goat diets in rangelands, and Israel and Texas share similar problems of brush encroachment. Therefore, our studies were intricately associated. Apart from intense e-mail exchanges, Dr Landau visited at TAES in February 2007 and Dr Walker visited at ARO in July 2007. Dr Walker was actively involved in tutoring the Israeli Ph.D. student Tzach Glasser by hosting him (on a 3-month Baron de Hirsch-funded visit) at San Angelo and facilitating his stay at Texas A&M University (College Station). Methods developed at ARO (fecal NIRS calibrated with the bite-count method) are being adopted at TAES and methods developed at TAES (such as the statistical analyses applied to NIRS calibration scores) are now routine at ARO. Apart from nine refereed papers already published or submitted to be published jointly, our cooperation triggered a Fecal NIRS session at the annual conference of the Society for Range Management in Reno, NV, February 2007, chaired by Dr. Walker. The best evidence of the fruitful collaboration between the two teams is the continuation program submitted to BARD in September 07.

Patent Summary (numbers)

Tatent Sammary (numbers)					
	Israeli inventor only	US inventor only	Joint IS/US inventors	Total	
Submitted					
Issued					
(allowed)					
Licensed					



#### **Abstract**

**Background**. Brush encroachment constitutes a serious problem in both Texas and Israel. We addressed the issue of efficacy of livestock herbivory - in the form of goat browsing - to change the ecological balance to the detriment of the shrub vegetation. Shrub consumption by goats is kept low by plant chemical defenses such as tannins and terpenes. Scientists at TAES and ARO have developed an innovative, cost-effective methodology using fecal Near Infrared Spectrometry to elucidate the dietary percentage of targeted, browse species (terpene-rich redberry and blueberry juniper in the US, and tannin-rich *Pistacia lentiscus* in Israel) for a large number of animals.

The original research objectives of this project were: 1. to clarify the relative preference of goat breeds and the individual variation of goats within breeds, when consuming targeted brush species; 2. to assess the heritability of browse intake and validate the concept of breeding goat lines that exhibit high preference for chemically defended brush, using juniper as a model; 3. to clarify the relative contributions of genetics and learning on the preference for target species; 4. to identify mechanisms that are associated with greater intake of brush from the two target species; 5. to establish when the target species are the most vulnerable to grazing. (Issue no.5 was addressed only partly.)

Major conclusions, solutions, achievements: Both the Israel and US scientists put significant efforts into improving and validating the technique of Fecal NIRS for predicting the botanical composition of goat diets. Israeli scientists validated the use of observational data for calibrating fecal NIRS, while US scientists established that calibrations could be used across animals differing in breed and age but that caution should be used in making comparisons between different sexes. These findings are important because the ability to select goat breeds or individuals within a breed for maximal efficiency of brush control is dependent upon accurate measurement of the botanical composition of the diet. In Israel it was found that Damascus goats consume diets more than twice richer in P. lentiscus than Mamber or Boer goats. In the US no differences were found between Angora and Boer cross goats but significant differences were found between individuals within breeds in juniper dietary percentage. In both countries, intervention strategies were found that further increased the consumption of the chemically defended plant. In Israel feeding polyethylene glycol (PEG, MW 4,000) that forms high-affinity complexes with tannins increased P. lentiscus dietary percentage an average of 7 percentage units. In the US feeding a protein supplement, which enhances rates of P450-catalyzed oxidations and therefore the rate of oxidation of monoterpenes, increased juniper consumption 5 percentage units. However, the effects of these interventions were not as large as breed or individual animal effects. Also, in a wide array of competitive tannin-binding assays in Israel with trypsin, salivary proteins did not bind more tannic acid or quebracho tannin than non-specific bovine serum albumin, parotid saliva did not bind more tannins than mixed saliva, no response of tannin-binding was found to levels of dietary tannins, and the breed effect was of minor importance, if any. These fundings strongly suggest that salivary proteins are not the first line of defense from tannin astringency in goats.

In the US relatively low values for heritability and repeatability for juniper consumption were found (13% and 30%, respectively), possibly resulting from sampling error or non-genetic transfer of foraging behavior, i.e., social learning. Both alternatives seem to be true as significant variation between sequential observations were noted on the same animal and cross fostering studies conducted in Israel demonstrated that kids raised by Mamber goats showed lower propensity to consume *P. lentiscus* than counterparts raised by Damascus goats.

**Implications:** The demonstration of differences in breeds and individuals within breeds, as well as intervention strategies, and maternal, non-genetic effects on the propensity of goats to consume chemically-defended shrubs provide invaluable knowledge for the implementation of environment-friendly procedures to control terpene-rich juniper (US) and tannin-rich *P. lentiscus* (IL) encroachment.



#### **Achievements**

#### Fecal NIRS to elucidate dietary botanical composition

The ability to select goat breeds or individuals within a breed for maximal efficiency of brush control is dependent upon accurate measurement of the botanical composition of the diet. Pre-BARD fecal NIRS calibrations of dietary browse percentages established with confined goats fed with three different browse species (including *P. lentiscus* and *Phyllirea latifolia*, the two main browse sources in this BARD project) did not validate well with direct observations of the dietary composition of goats throughout whole grazing days. Principal component analysis also showed that fecal spectra from confined and ranging animals could not constitute one population. The first step of the project in Israel was, therefore, to establish the first fecal NIRS calibrations, based on direct observations, encompassing 90 observation days with Mamber, Damascus, and Boer goats, encompassing more than 200,000 bites in four seasons. These calibrations — representing the first fecal NIRS system based on observations ever published - were yearly upgraded and always found to be precise, accurate, and robust to field conditions. They constitute the cornerstone of the project in Israel.

In parallel, it was found in the US that internal validations of fecal NIRS-predicted juniper in the diet had a high precision, but the precision of independent validations was low. Spectral differences were affected by diet, sex, breed, and age, but diet was the largest source of variation, accounting for 95% of the variation in predictions from internal calibrations and 51% of the variation in independent validations. When two complete mixed rations differing in juniper content were fed to goats of differing breed, gender, and age, predictions from independent calibrations readily detected differences in the dietary percentage of juniper, and the predicted differences were similar to the actual differences. Hence, the US fecal near-infrared reflectance spectroscopy predictions of dietary botanical composition were considered an interval scale of measurement and used to detect heritability and repeatability of dietary juniper in the US.

#### Breeds, within-breed variability, heritability, and social learning

In Israel, *P. lentiscus* formed 13% of the DM ingested by Damascus goats, but only 5%, for the Mamber and Boer goats, with Damascus goats ingesting diets 23% richer in tannins, suggesting this breed as preferred candidate for the control of *P. lentiscus* encroachment.

The US found significant variability between individuals within breeds in the amount of juniper that goats consumed. Heritability for juniper consumption was estimated at 13% and repeatability was estimated to be 30%, *i.e.*, lower than previously calculated heritability and repeatability values for chemically defended plants in the US. The difference between the results of this study and previous



studies could be caused by sampling error or non-genetic transfer of foraging behavior, i.e., social learning. Further BARD research confirmed that both hypotheses were true. When high and low juniper consuming Angoras were sampled twice per week for two years, significant variation existed between sequential observations on the same animal. Also, non-genetic-transfer of foraging behavior was evidenced in a cross-fostering experiment between Damascus and Mamber does, and their kids. Before being turned to grazing with their biological or fostering mothers, Damascus and Mamber kids showed equal propensity to ingest tannin-rich *P. lentiscus* or low-tannin *P. latifolia* when given alone, and equal dietary percentage of the two species when given together. After the grazing period, the kids "educated" by Damascus does or by artificial milk-dispensers consumed 50% more *P. lentiscus* – under controlled conditions - than kids educated by Mambers.

Hence, the importance of environmental or learning effects was demonstrated in Israel and the presence of large permanent environmental effects were also seen in the US heritability calculations. Further research is warranted to determine how to best use environmental effects to increase consumption of targeted browse and how such effects may be influencing heritability estimates.

### Physiological adaptations

Can an underlying physiological difference be found between goats with high and low genetic merit for juniper consumption (US) or with high and low propensity to consume *P. lentiscus* (IL)?

As monoterpenes and barbiturates are metabolized by the same pathway, goats with a greater ability to metabolize barbiturates presumably would have a greater ability to metabolize monoterpenes. When we measured the rate of drug metabolism as measured pentobarbital sleep time in goats with high or low genetic merit for juniper consumption, there was no correlation between sleeping time and index of genetic merit, but the goats with the highest predictions of genetic merit had the longest sleeping times. The slower rate of hepatic metabolism of pentobarbital seen with the high juniper consuming group suggests that a slow clearance rate of circulating monoterpenes is potentially beneficial to increasing juniper intake. Our original hypothesis was that a higher rate of clearance would reduce the pharmacological impact of the circulating parent compound. An equally valid hypothesis that could explain our results is that the toxic effects of monoterpenes are not exerted by the parent compound but rather by a bioreactive intermediate formed through hepatic phase I oxidation.

In Israel, it was hypothesized that specific salivary proteins, sourcing in the parotid salivary gland, bind to dietary tannins, allowing for higher ingestion of tannin-rich browse. Saliva was sampled from the cannulated parotid or from the oral cavity in Damascus, Boer, and Mamber goats fed diets



with or without *P. lentiscus*. A competitive colorimetric test involving trypsin as target protein and quebracho tannin or tannic acid as tannins, showed that parotid saliva does not bind tannins more than mixed saliva; mixed and parotid saliva proteins do not bind more tannin than the nonspecific bovine serum albumin; saliva from Damascus goats does not bind more tannins than saliva from Mambers; and tannin-binding activity is not correlated to diet. All these findings strongly suggest that salivary proteins are not the first line of defense from tannin astringency in goats.

Interestingly, the BARD results challenged scientific paradigms in the two countries.

#### **Interventions**

In Israel feeding polyethylene glycol (PEG, MW 4,000) that forms high-affinity complexes with tannins increased *P. lentiscus* dietary percentage from 10 to 17% but PEG supplementation could only partly bridge breed-related differences in the propensity to consume *P. lentiscus*.

In the US feeding a protein supplement increased juniper consumption 5 percentage units.

In both countries, intervention strategies increased the consumption of the chemically defended plant, but the effects of these strategies were not as large as the breed or individual animal effects.

**Implications**: The results outlined above exposed additional knowledge gaps that should be addressed to increase the use of goats for brush management. The discovery in both countries of differences in breeds and individuals within breeds as well as intervention strategies to increase consumption of chemically defended plants, begs the question – will goats with a higher preference for a target plant be more effective in controlling that plant than other goats? - that we have proposed to address in our continuation project.

#### **Publications**

- Campbell, E.S., C.A. Taylor, J.W. Walker, C. J. Lupton, D.F. Waldron, S.Y. Landau. (2007). Effects of supplementation on juniper intake by goats. *Rangeland Ecology and Management*, 60: 588-595.
- Glasser, T., Landau, S., Ungar, E.D., Perevolotsky, A., Dvash, L., Muklada, H., Kababya, D., Walker, J.W. (2008). A fecal NIRS-aided methodology to determine goat dietary composition in a Mediterranean shrubland. *Journal of Animal Science*, doi:10.2527/jas.2006-817 (in print)
- Walker, J.W., Campbell, E.S., Lupton, C.J., Taylor, Jr., C.A., Waldron, D.F., Landau, S.Y. (2007).
  Contribution of breed, sex, and age to variation and predictive ability of near-infrared reflectance spectra of goat feces. *Journal of Animal Science*, 85:518-526.



## **Published papers**

- Campbell, E.S., C.A. Taylor, J.W. Walker, C. J. Lupton, D.F. Waldron, S.Y. Landau. (2007). Effects of supplementation on juniper intake by goats. *Rangeland Ecology and Management*, 60: 588-595.
- Glasser, T., Landau, S., Ungar, E.D., Perevolotsky, A., Dvash, L., Muklada, H., Kababya, D., Walker, J.W. (2008). A fecal NIRS-aided methodology to determine goat dietary composition in a Mediterranean shrubland. *Journal of Animal Science*, doi:10.2527/jas.2006-817 (in print)
- Walker, J.W., Campbell, E.S., Lupton, C.J., Taylor, Jr., C.A., Waldron, D.F., Landau, S.Y. (2007).
  Contribution of breed, sex, and age to variation and predictive ability of near-infrared reflectance spectra of goat feces. *Journal of Animal Science*, 85:518-526

#### Manuscript submitted for publication

Glasser, T., Landau, S., Ungar, E.D., Muklada, H., Perevolotsky, A., Dvash, L., Muklada, H., Kababya, D., Walker, J.W. Foraging selectivity of three goat breeds in a Mediterranean shrubland. Submitted to *Applied Animal Behaviour Science* 

# Manuscripts in preparation

- Campbell, E.S., Taylor, C.A., Jr., Walker, J.W., Lupton, C.J., Waldron, D.F., Landau, S.Y..Effect of monoterpene concentration on juniper consumption in goats To be submitted to *The Journal of Chemical Ecology*
- Glasser, T., Landau, S., Ungar, E.D., Muklada, H., Perevolotsky, A., Dvash, L., Muklada, H., Kababya, D., Walker, J.W. Effect of polyethylene-glycol on foraging selectivity of three goat breeds in Mediterranean shrubland: A Fecal NIRS-aided study. To be submitted to *Small Ruminant Research*
- Glasser, T., Landau, S., Perevolotsky, A., Muklada, H., Ungar, E.D., Walker, J.W. Breed and maternal effects on the intake of tannin-rich browse by domestic goats (*Capra hircus*). To be submitted to *Applied Animal Behaviour Science*
- Hanovitz-Ziony, M., Gollop, N., , Muklada, H., Brener, S., Ungar, E.D, Landau, S. Walker, J.W. Salivary proteins are not the first defense line against dietary tannin astringency in goats. To be submitted to *The Journal of Chemical Ecology*



- Waldron, D.F., Taylor, C.A., Jr., Walker, J.W., Campbell, E.S., Lupton, C.J., Willingham, T.D. Landau, S.Y.. Heritability of juniper consumption in goats. To be submitted to *Journal of Animal Science*
- Walker, J.W. Campbell, E.S., Waldron, D.F., Taylor, C.A., Jr., Lupton, C.J., Landau, S.Y.. Periodicities in juniper consumption in goats. To be submitted to *Applied Animal Behaviour Science*

#### M.Sc. and Ph.D. Theses

- Hanovitz-Ziony, M. (2008). Does saliva represent the first line of defense against dietary tannin astringency in goats? M.Sc. Thesis. The Faculty of Agriculture and Environmental Quality, the Hebrew University, Rehovot, Israel.
- Glasser. T. (2008). Breed and mother effects on the intake of *Pistacia lentiscus* in Mediterranean shrubland by goats: A Fecal NIRS-aided study. Ph.D. Thesis. The Faculty of Agriculture and Environmental Quality, the Hebrew University, Rehovot, Israel.
- Lovett, T.D. (2007). Mohair production and quality in two divergent selection lines of Angora goats. M.S. Thesis. Angelo State Univ., San Angelo, TX

#### Abstracts presented at international research conferences

- Walker, J.W., Landau, S.Y. (2007). Predicting botanical composition of ruminant diets with fecal Near Infrared Reflectance Spectroscopy. 60th Annual Meeting Americal Society Range Management, 10-16 February 2007, Reno (NV, USA).
- Landau, S.Y., Glasser, T.A., Ungar, E.D., Perevolotsky, A., Muklada, H., Walker, J.W. (2007).
  Fecal NIRS for the study of goat nutrition in a shrubland rich in *Pistacia Lentiscus*. 60th
  Annual Meeting Americal Society Range Management, 10-16 February 2007, Reno (NV, USA).
- Walker, J., Campbell, E.S., Waldron, D.F., Lupton, C.J., Taylor, C.A., Jr., Engdahl, B.S., Landau, S. (2008). Periodicities in consumption of chemically defended plants. Proc. 61st Annual Meeting Americal Society Range Management, 25-31 January 2008, Lexington (KY, USA).
- Frost, R., Campbell, E.S., Brewer, T.K., Mosley, J.C., Taylor, C.A., Walker, J., Waldron, D.F., Lupton, C.L. (2008). Pharmacokinetic differences in high and low juniper consuming



goats. 61st Annual Meeting American Society Range Management, 25-31 January 2008, Lexington (KY, USA).

### **Chapters in Book (in press)**

Walker, J.W., Whitworth, W.R., Campbell, E., Taylor, C.A., Jr., Scott, C.B., Lupton, C.J., Kott, R., Surber L., and Landau S.. Fecal NIRS for predicting botanical Composition of Herbivore diets. - In: Fecal Near-Infrared Reflectance Spectroscopy for Determining Attributes of Herbivores and Their Diets: The State of the Science. John W. Walker and Doug Tolleson editors. Texas AgriLife Research Bulletin. College Station TX, USA.

Landau, S., Glasser, T., Ungar, E.D., Perevolotsky, A., Dvash, L., Muklada, H., Kababya, D., and Walker, J.W. Fecal NIRS with bite counts: A methodology to determine the botanical and chemical composition of diets consumed by goats in a Mediterranean shrubland - In: Fecal Near-Infrared Reflectance Spectroscopy for Determining Attributes of Herbivores and Their Diets: The State of the Science. John W. Walker and Doug Tolleson editors. Texas AgriLife Research Bulletin. College Station TX, USA.